REMARKS

Claims

A typographical error is corrected by the amendment of Claim 25 so that "*ne* detergent" is replaced by "*one* detergent".

Claim Rejections:

In the June 3, 2003 Office Action, the Examiner rejected Claims 1-8 and 14 and objected to Claims 15, 18, 30, 42 and 59. The remaining Claims (9-13, 16-17, 19-29, 31-41, and 43-58) are not discussed in the June 3, 2003 Office Action.

In the June 3, 2003 Office Action the Examiner states the objected to claims would be allowable if rewritten as independent claims and, accordingly, the applicants submit additional claims to respond to this suggestion. However, the applicants respectfully assert that none of the claims are either anticipated or obvious and assert that all of Claims 1-64 are allowable.

The claimed invention

The present invention is drawn to a method "to detect ATP in a sample by reducing the steps of cell lysis, endogenous ATPase inhibition, and substrate and luciferase addition *to a single step*." Specification, page 4, lines 10-12 (*emphasis added*). This method differs from the prior art where cell lysis and luciferase addition was conducted in a stepwise manner." Specification, page 3, lines 29-30. "[C]urrent assays that use luminescence to detect ATP are handicapped by the need for successive, time-consuming steps." Specification, page 4, lines 6-7. All of the claims require that cell lysis, endogenous ATPase inhibition, and substrate and luciferase addition occur in a single step.

The single step assay of the present invention is possible because applicants have discovered a class of luciferases which are stable to enzymatic degradation. Hence, claim 1 requires that the "reagent composition is capable of maintaining at least

about 30% activity, as measured by luminescence after the reagent composition is combined with the sample, for at least one hour compared to the reagent composition's activity just after the luciferase is combined with the one or more detergents."

35 U.S.C. § 102

Claims 1-8 and 14 are rejected under 35 U.S.C. § 102(b) over Simpson et al. The Office Action states, "Simpson et al. disclose a bioluminescent method for assaying of ATP in a sample by contacting a reagent composition comprising a cationic surface active agent to the sample, adding an amount of non-ionic surface active agent, *followed by* determining the ATP released into the sample using the luciferase-luciferin reagent." (*Citing,* Simpson, column 6, lines 53-68 and column 7-8; lines 1-18; *emphasis added*). Whereas Simpson requires the step-wise addition of reagents, the present invention adds a single reagent composition including both detergents and enzyme in a single step. For example, claim 1 recites, "adding to the sample a reagent composition comprising one or more detergents and a luciferase."

Simpson teaches the step-wise, separate addition of the ATP releasing agent (cationic detergent) and neutralizing agent (nonionic detergent) throughout their specification. See e.g. Simpson, column 3, lines 10-15 & 32-50; column 4, lines 1-4. Further, Simpson claims stepwise addition detergent followed by luciferase-luciferin reagent ("and *thereafter* determining the ATP released into the sample using the luciferase-luciferin reagent," Simpson, claim 1, line 58-60 (*emphasis added*)). Similarly, Claim 5 of Simpson teaches the separate, step-wise addition of the cationic surface active agent to release ATP (step (b)), the nonionic surface active agent to neutralize the cationic surface active reagent (step (c)), and the addition of firefly luciferin-luciferase (step (e)). Simpson, Column 7, line 14 to Column 8, line 11. Nowhere does Simpson teach or claim a reaction in which cationic detergent, nonionic detergent, and luciferase-luciferin reagents are simultaneously added to the sample to be assayed.

The Examiner also contends that Simpson discloses a reagent composition comprising both a cationic surface active agent and a non-ionic surface active neutralizing agent. (*citing* Simpson, Column 3, line 16-19). However, the cationic surface active agent and non-ionic surface active agent are added to the assay in separate steps. The assay is to be performed by contacting the cellular ATP source "with ATP releasing agent and contacting *the resultant solution* with a neutralizing agent" Simpson, column 3, lines 13-14. (With the releasing agent preferably a cationic surface agent and the neutralizing agent a non-ionic surface active agent. Simpson, column 3, lines 16-19.) Again, Simpson fails to set forth the one step addition of cationic detergent, nonionic detergent, and luciferase-luciferin reagents.

The Examiner contends that Simpson also discloses combinations of cationic surface active agents, ionic detergents, and non-ionic detergents. *Citing* Simpson, column 2, lines 1-68. But Simpson only refers to these agents in conjunction with discussions of the problems inherent in their use. Simpson notes that cationic surface active agents reduce the precision of the ATP assay (Simpson, column 2, lines 30-40) and that a mixture of ionic detergents requires dilution prior to performing the assay, and that non-ionic detergents may distort the kinetics of the luciferase assay (Simpson, column 2, lines 41 to column 3, line 7). In view of the problems with the use of detergents, Simpson is better said to teach away from the addition of cationic surface active agents, ionic detergents, and non-ionic detergents in a single step of an ATP assay.

The Examiner contends that Simpson anticipates the present application when it teaches the use of EDTA (column 3, lines 8-68 and column 4, lines 1-68). The Examiner finds anticipation when Simpson et al. teach the use of a cationic detergent at the concentration of 0.1% (w/v) or greater and the use of the detergents used in the present application's claim 14. However, again, in Simpson EDTA and these detergents are not simultaneously added to the sample with luciferase in a single step.

The Examiner concedes that the present application's Claim 1 limitation that "the reagent composition is capable of maintaining at least about 30% activity, as measured

by luminescence after the reagent composition is combined with the sample, for at least one hour" is not disclosed by Simpson. However, the Examiner contends that this element's disclosure is "an inherent property of the reaction mixture in the ATP assay disclosed by the Simpson et al. reference because Simpson et al. disclose methods that use the same starting materials and reaction conditions claimed in the present invention." June 3, 2003 Office Action, Page 3 ¶ 2.

Anticipation under 35 U.S.C. § 102 requires that each and every element be set forth in a single prior art reference. MPEP § 2131 (8th ed. 2001); *Verdegaal Bros. v. Union Oil Co. of Calif.,* 814 F.2d 628, 631 (Fed. Cir. 1987). The express, implicit or *inherent* disclosures of a prior art reference may be relied upon for a rejection of claims under 35 U.S.C. § 102. MPEP § 2112 (8th ed. 2001); *In re Napier*, 55 F.3d 610, 613 (Fed. Cir. 1995). However, the Examiner must provide a basis in fact or technical reasoning to reasonably support the contention that the allegedly inherent characteristic *necessarily* flows from the teachings of the prior art. MPEP § 2112 (8th ed. 2001); *Ex parte Levy,* 17 U.S.P.Q.2d 1461, 1464 (Bd. Pat. App. & Inter. 1990). It is not enough that a result or characteristic may occur or be present in the prior art to establish inherency. MPEP § 2112 (8th ed. 2001); *In re Rijckaert,* 9 F.3d 1531, 1534 (Fed. Cir. 1993).

Simpson does not explicitly disclose any data on a luciferase reaction monitored for more than a few seconds. (*See, e.g.,* Fig. 2 Simpson.) Moreover, the reaction conditions are different in Simpson from the present application because Simpson teaches the multi-step addition of reagents in its ATP assay. Therefore, it is not clear that a process that maintains 30% of the luciferase activity after one hour *necessarily* flows from the teachings of Simpson. Thus, Simpson does not anticipate any of the claims of the present application.

Furthermore, Examples 3 and 7 of the present application suggest that Simpson does not anticipate the present invention. Example 3 of the present application is similar to Example 1 of Simpson. Both examples disclose the degree of luciferase stability in the presence of cationic detergent after various times and in various

concentrations of the cationic detergent. Simpson's Figure 1 demonstrates that the cationic detergent benzethonium chloride cause a decrease in the luciferase enzymes activity (measured by light intensity decay versus starting light intensity) of up to 600% per minute (at 0.05% benzethonium chloride w/v). The present application discloses assay conditions where a wild-type luciferase enzyme ("LucPpy" in Table J) in the presence of 0.02% (w/v) of the cationic detergent dodecyltrimethylammonium bromide (DTAB) retains 99.53% of its activity (as measured by luminescence generated in response to ATP) after 34 minutes. Specification, page 51, Table J.

In addition, using a genetically engineered luciferase ("LucPpe2m146" in Table J), the present application discloses assay conditions in which a substantial amount of the luciferase activity and luminescence is retained after addition of the cationic detergent DTAB. Specification, page 51, Table J.

Similarly, the present application's Example 7 demonstrates that luciferase stabilization is not achieved in Simpson's assay. The reaction mixture disclosed in the present application maintains luciferase activity even in the presence of DTAB over the course of 325 minutes (Specification, Page 60, Table Q, which discloses luciferase activity half-lives ranging from 1.4-14.2 hours) while Simpson's Figure 1 demonstrates that the cationic detergent benzethonium chloride causes light decay of up to 600% per minute (a luciferase activity half-life of less than one minute). Thus, Simpson does not teach an assay with the properties of enhanced luciferase stability disclosed in the present application and Simpson does not anticipate the present application's claim elements requiring the maintenance of at least about 30% activity for at least one hour.

35 U.S.C. § 103

The Examiner alternatively contends that Simpson renders the present application's claims 1-8 obvious under 103(a). To make a *prima facie* case of obviousness, the Examiner must first show there is suggestion or motivation, either in the references themselves or in the knowledge generally available to one of ordinary skill in the art, to modify the reference or to combine reference teachings. Second, there must be a reasonable expectation of success. Finally, the prior art reference (or

references when combined) must teach or suggest all the claim limitations. Importantly, the teaching or suggestion to make the claimed combination and the reasonable expectation of success must both be found in the prior art, not in applicant's disclosure. MPEP §2143 (8th ed. 2001); *In re Vaeck*, 947 F.2d 488, 20 USPQ2d 1438 (Fed. Cir. 1991).

Simpson fails to suggest the combined addition of all the detergents and luciferase reagents. Instead, as discussed above, Simpson teaches away from such a combined addition step.

The present invention is not obvious because it discloses an assay that allows the addition of detergents and luciferase in a single step. The disclosed one-step assay is possible because the reagent composition disclosed has properties of enhanced stability. In addition, although luciferase functions as an ATPase, in the present application's reagent composition luciferase is resistant to the effects of the ATPase inhibitor also present in the reagent composition. Specification, page 4, line 23-27.

Nothing in Simpson suggests that the combination of cell lysis, the inhibition of endogenous ATPase, and substrate and luciferase addition to a single step will be successful. Simpson does state that the "careful manipulation of the ratio of one surface active agent to the other...could limit (but not eliminate) the decay" of the luciferase reaction rate. However, Simpson follows by warning that these agents reduce the assay precision. Simpson, column 2, lines 35-40. Thus, the prior art does not offer a reasonable expectation of success for a modification of the prior art constituting the invention claimed in the present application.

Claims 9-13, 16-17, 19-29, 31-41, and 43-58:

There is no rejection of record for these claims. To the extent that the examiner intended to reject these claims under 102(b) over Simpson, the Office Action fails to provide any explanation of how Simpson meets the various claimed limitations. As mentioned above, all of the claims, including claims 9-13, 16-17, 19-29, 31-41, and 43-58, require the single step addition of detergent and luciferin-luciferase reagent. As this

element is not provided by Simpson, Claims 9-13, 16-17, 19-29, 31-41, and 43-58 are also allowable.

Objections to Claims:

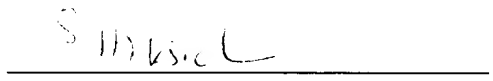
The Examiner has objected to Claims 15, 18, 20, 42 because they are based on rejected independent claims. The Examiner states these claims would be allowable if amended to independent claim formats. Accordingly, we present the new Claims numbers 60-64 which restate the objected to claims in an independent claim format.

However, the applicants respectfully submit that, because none of the Claims are anticipated or obvious, Claims 15, 18, 20, 42 should be allowed as originally written.

SUMMARY

The applicants believe that currently pending Claims 1-64 are patentable. The applicants respectfully request the Examiner grant early allowance of this application. The Examiner is invited to contact the undersigned attorney for the applicants via telephone if such communication would expedite this application.

Respectfully submitted,



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